

AMENDMENTS TO THE CLAIMS

1. (Currently Amended) Method of purifying an antibody, ~~preferably an IgG antibody,~~ comprising the steps of:
 1. Purifying an antibody by means of protein A affinity chromatography wherein the protein A is a native protein A or a functional derivative thereof,
 2. loading the thus purified antibody comprising antibody aggregate and protein A or protein A derivative onto an ion ~~exchange~~exchanger material under conditions which allow of binding of the contaminating protein A or its functional derivative to the ion exchanger ~~material~~ and which conditions further allow of resolution in the flow-through of antibody aggregates from antibody monomer which monomer is not complexed with protein A or protein A derivative ~~by means of fractionation of the flow-through,~~ and further
 3. fractionating the flow-through of step 2) and harvesting from the flow-through ~~of the ion exchanger~~ at least one antibody monomer fraction having both reduced contents of protein A or protein A derivative and further reduced contents of antibody aggregate as compared to the composition of antibody as loaded onto the ion ~~exchange~~exchanger material before by fractionating or splitting the antibody peak of the flow-through into at least two fractions and wasting the tail fraction.
2. (Original) Method according to claim 1, characterized in that the protein A is a recombinant protein A that is engineered such as to allow of single-point attachment to a column material.
3. (Original) Method according to claim 2, characterized in that the recombinant protein A comprises a cysteine in its amino acid sequence.
4. (Original) Method according to claim 3, characterized in that the cysteine is comprised in a segment that consists of the last 30 Amino acids of the C-terminus of the amino acid sequence of the recombinant protein A.
5. (Cancelled)

6. (Currently Amended) Method according to claim 31, characterized in that the protein A or its functional derivative is reduced to a concentration of <1ng/mg IgG in the flow-through of the ion exchanger.
7. (Currently Amended) Method according to claim 31, characterized in that the monomericity of the antibody harvested is at least 99% ~~and is achieved by fractionating the antibody peak of the flow through into at least two fractions and wasting the tail fraction.~~
8. (Currently Amended) Method according to claim 31, characterized in that the antibody is a monoclonal antibody, ~~preferably an IgG antibody wherein the IgG antibody may be chimeric or CDR-grafted IgG antibody.~~
9. (Currently Amended) Method according to claim 31, characterized in that the antibody is harvested from a cell culture prior to purifying the antibody by means of protein A affinity chromatography.
10. (Currently Amended) Method according to claim 31, characterized in that the antibody is harvested from a mammalian cell culture.
11. (Currently Amended) Method according to claim 31, characterized in that the antibody that is to be purified by means of protein A affinity chromatography is not treated as to inactivate proteases, ~~preferably is not in admixture with at least one protease inhibitor.~~
12. (Cancelled)
13. (Currently Amended) Method of purifying a product protein, comprising the steps of:
 1. loading a solution comprising product protein which product protein comprises monomeric and aggregated forms of said protein onto an ion ~~exchange~~exchanger ~~material~~ under conditions which allow of resolution in the flow-through of said product protein aggregates

from said product protein monomer which monomer ~~preferably~~ is not further complexed with a second protein ligand, ~~by means of fractionation of the flow-through~~ and further 2. fractionating the flow-through of step 1) and harvesting from the flow-through ~~of the ion exchanger~~ at least one product protein monomer fraction having reduced contents of product protein aggregate as compared to the composition of product protein loaded onto the ion ~~exchange~~ exchanger material for purification by fractionating or splitting the product protein peak of the flow-through into at least two fractions and wasting the tail fraction.

14-15. (Cancelled)

16. (Currently Amended) Method according to claim ~~15~~1, characterized in that at least one buffer is used for loading and rinsing the ion exchanger which at least one buffer coming off the ion exchanger is constituting the flow-through comprising the product protein peak.

17. (Original) Method according to claim 16, characterized in that the pH of said buffer is set at a pH which is the pI or average pI of the product protein monomer sought to be purified in the range of ± 0.5 pH units around said pI.

18. (Currently Amended) Method according to claim 16, characterized in that the pH of said buffer is set at a pH different from the pI or average pI of the product protein monomer sought to be purified and which pH further vests the product protein monomer with a surface charge which charge leads to ionic attraction in between product protein monomer and the charged groups of the ion ~~exchange~~ exchanger material when exposed to or submerged in said buffer.

19. (Currently Amended) Method according to claim 18, characterized in that in case of a cation exchanger, the pH of the buffer is set at a value below the average pI of the product protein monomer sought to be purified, ~~preferably set at a value of from 0.5 to 3 pH units below said average pI.~~

20. (Currently Amended) Method according to claim 18, characterized in that in case of an anion exchanger, the pH of the buffer is set at a value above the average pI of the product protein monomer sought to be purified, ~~preferably set at a value of from 0.5 to 3 pH units above said average pI.~~
21. (Currently Amended) Method according to claim 13, characterized in that said conditions are non-binding conditions as regards binding of the product protein monomer to the ion exchanger ~~material~~ such as that consequently more than 70% (w/w), ~~more preferably more than 80% (w/w)~~ of the product protein loaded onto the ion ~~exchange~~exchanger ~~material~~ can be recovered in the flow-through from the ion ~~exchange~~exchanger ~~material~~.
22. (Cancelled)
23. (New) Method according to claim 8, characterized in that the monoclonal antibody is an IgG antibody wherein the IgG antibody may be chimeric or CDR-grafted IgG antibody.
24. (New) Method according to claim 23 above, characterized in that the IgG antibody is a chimeric or a CDR-grafted IgG antibody.
25. (New) Method according to claim 19, wherein the pH of the buffer is set at a value of from 0.5 to 3 pH units below said average pI.
26. (New) Method according to claim 20, wherein the pH of the buffer is set at a value of from 0.5 to 3 pH units above said average pI.